# REGULAR ARTICLE

# New agricultural practices in the Loess Plateau of China do not reduce colonisation by arbuscular mycorrhizal or root invading fungi and do not carry a yield penalty

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Abstract Agricultural practices aimed to reduce soil erosion and improve crop yield have been suggested to influence the activity of arbuscular mycorrhizal (AM) and root pathogenic fungi. We conducted a two-year field survey to investigate the effect of recently introduced agricultural practices on crop yield, AM colonisation and percentage isolation of root-invading fungi on the heavily eroded Loess Plateau of China. A rotation of maize (Zea mays L.), winter wheat (Triticum aestivum L.) and soybean (Glycine max L.) replaced monoculture of winter wheat. No-tillage (NT), and return of previous crop residues to the field in tilled (CTR) and non-tilled

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(NTR) systems replaced conventional tillage (CT). Yield, biomass and phosphorus content of the crops showed similar trends. Residue application increased yields of maize and soybean independent of tillage treatment in 2004, but only under CT in 2005. CT slightly increased maize yield. Neither residue application nor tillage treatment affected yield of wheat. None of the treatments influenced total percent isolation of root-invading fungi from wheat roots. The increase of some individual pathogenic fungi in NT did not translate into reduction of yield by disease. Importantly, the recommended practices did not have a penalty on yield while maintaining high levels of AM colonisation.

Keywords Arbuscular mycorrhiza . Root-invading fungi . Tillage . Residue application . Phosphorus uptake

# Introduction

The Loess Plateau is the area with highest soil erosion in China and also amongst the highest in the world (Fu [1989;](#page-9-0) Liu [1999\)](#page-9-0). About  $16.4\times10^8$  t of soil is lost from this area to the Yellow River each year (Shi and Shao [2000\)](#page-10-0), resulting in poor soil fertility, desertification and formation of steep slopes and valleys. Intensive tillage was thought to be one of the main reasons for erosion in this area. These conventional tillage practices consisted of three deep ploughs each year, continuous monoculture, removal of all residues from the field at harvest for fuel or animal feed and leaving the field without crop cover for 3–8 months (Huang et al. [2008](#page-9-0)). All these activities have been shown to accelerate soil erosion and reduce crop production (Liu [1999\)](#page-9-0).

Conservational agricultural practices have been undertaken in Xifeng, Gansu Province, located on the Loess Plateau of China, since 2001 with the aim of improving crop production and reducing soil erodability. These practices consisted of no tillage (NT), no tillage and application of residues of the previous crop (NTR), or conventional tillage (CT) and application of residues of the previous crop (CTR). In Xifeng, the typical agricultural system is rainfed cereal production and 97% of householders are farmers (Hardiman et al. [1990](#page-9-0)).

The benefits of using no tillage and applying crop residues as surface mulch to increase soil organic carbon (C) content, improve soil physical properties and water infiltration and reduce soil erosion are widely recognised in many parts of the world (Triplett Jr and Dick [2008\)](#page-10-0), including Gansu province (Huang et al. [2008](#page-9-0)). The easily oxidised organic C increased significantly under no tillage with application of residues of the previous crop, compared with that under the conventional treatment, which suggests that the newly-introduced practices may be successful in increasing soil structural stability (Luo et al. [2005](#page-9-0)), which is closely correlated with crop yield. Indeed, reduced tillage on the Loess Plateau has been shown to increase the proportion of water stable aggregates in the size range of 0.25 mm to 2 mm (Luo et al. [2005\)](#page-9-0) and AM fungi may contribute to this process.

Symbiotic associations between plants and AM fungi are ubiquitous in terrestrial plant communities, including important agricultural crops. AM symbiosis can enhance plant mineral nutrient uptake, particularly P, improve tolerance to water stress, resistance to disease and improve soil structural stability and hence help reduce soil erosion (Smith and Read [2008](#page-10-0)). External hyphae of AM fungi in soil can contribute to the formation of macroaggregates  $(>0.25$  mm diameter) and are positively correlated with aggregate stabilization in natural systems (Tisdall and Oades [1979\)](#page-10-0). In some systems adoption of reduced tillage has been shown to increase colonisation of crop roots by arbuscular mycorrhizal (AM) fungi, with consequent increase in phosphorus (P) uptake (Evans and Miller [1988;](#page-9-0) Fairchild and Miller [1988](#page-9-0); McGonigle and Miller [1996](#page-9-0)).

Conversely, there are reports of increases in root diseases associated with minimum or no tillage; e.g. in disease-suppressive soils in South Australia (Rhizoctonia solani; Roget [1995](#page-9-0); Rovira [1986](#page-9-0)) and in corn fields in Colorado, USA (Fusarium spp.; Skoglund and Brown [1988](#page-10-0)). Soil-borne diseases caused by rootinvading fungi, such as R. solani and Fusarium spp., generate important reductions in crop yield worldwide (Skoglund and Brown [1988](#page-10-0); Rovira [1986\)](#page-9-0). These fungi can survive many years in soil and have a wide range of host plants, so that diseases caused by them are very difficult to control (Pankhurst et al. [2002;](#page-9-0) Skoglund and Brown [1988\)](#page-10-0). Tillage and residue management (including stubble retention and return of residues to the field) have been suggested to depress root rot disease caused by R. solani in some agricultural systems (Pankhurst et al. [2002](#page-9-0)) but have contributed to an increase in disease severity in others (Roget [1995](#page-9-0); Rovira [1986](#page-9-0)).

The outcomes of the adoption of reduced or no tillage and crop residue retention on AM fungal colonisation and root disease incidence have not been investigated on the Loess Plateau. We hypothesised that AM colonisation and incidence of other rootinvading fungi would be increased under no tillage and would potentially also be affected by returning crop residues to the fields. We surveyed a long term ongoing field trial to investigate these hypotheses. We assessed the effects of conservational agricultural practices on AM colonisation in three crops during two years (2004 and 2005) and on the percentage isolation of root-invading fungi (mostly pathogenic) from wheat roots in 2004. Crop growth, P uptake and grain yield were also determined in order to set the work in context of overall agronomic management.

## Materials and methods

### Trial site

A long term trial was established at the Qingyang Loess Plateau Field Station of the College of Pastoral Agriculture Science and Technology of Lanzhou University, which lies in the middle of the Longdong Loess Plateau, Gansu Province, China (35°39′N, 107° 51′E) at an altitude of approximately 1,297 m.

<span id="page-2-0"></span>Table 1 Conditions at the experimental site Longdong Loess Plateau, Gansu Province, China

| Altitude (m)  | 1.297          | Frost free days per year       | 162         |
|---|----------------|--------------------------------|-------------|
| Mean temperature $(^{\circ}C)$                          | $8.0 - 10.0$   | Growing season (days per year) | 255         |
| Temperature range $(^{\circ}C)$                         | $-22.4 - 39.6$ | Annual rainfall (mm)           | $480 - 660$ |
| Annual mean accumulated temperature $(>5^{\circ}C)^{a}$ | 3.446          | Soil pH                        | $8 - 8.5$   |

<sup>a</sup> mean daily temperatures above  $5^{\circ}\text{C}$  accumulated over a year. Winter wheat requires above  $2,123^{\circ}\text{C}$  to finish all growth stages and produce significant yield; maize and soybean require above 1,900°C and 1,850°C (>10°C accumulated temperature) respectively

Environmental conditions are summarised in Table 1. The soil in this area is a sandy loam with low fertility, soil organic carbon below 1.70% and Olsen-P below 25 mg/kg (Olsen et al. [1954](#page-9-0)).

#### Experimental design

The trial was set up as a maize, winter wheat and soybean rotation system in 2001. There were four treatments: (1) conventional tillage (CT)—top soil (20 cm) tilled twice a year (September before wheat sowing and October after soybean harvest), without addition of residues; (2) conventional tillage with previous crop residue returned to the field after harvest (CTR); (3) no till (NT)—soil disturbed only when sowing by machine, without addition of residues; and (4) no till with residue returned (NTR)—sown as for NT and previous crop residue returned.

After harvest residues of soybean, winter wheat and maize were ground or cut into 5–10 cm fragments. All the soybean and wheat residues and 50% of the maize residues were returned to the field by laying them on the soil surface.

There were four replicate plots for each treatment, randomly arranged with a total of 16 plots for each phase (see below). The plot size was  $4 \text{ m} \times 14 \text{ m}$  $(56 \text{ m}^2)$  with one metre space between each plot.

Plant and rotation sequence

All crops were varieties sown by local farmers: maize (Zea mays L.cv Zhongdan No.2), winter wheat (Triticum aestivum L. cv Xifeng No.24) and soybean (Glycine max L. Merr. cv Fengshou No.12).

Crops were sown according to local practice. Maize was sown in April, with a 38 cm space between rows and 10 cm distance between plants, and harvested in late September each year. Winter wheat was sown immediately after the maize harvest with a 15 cm space between rows at a planting density of 187 kg/ha. After winter wheat had been harvested in June or July the following year, soybean was sown with a 25 cm space between rows and plants, and harvested in October. A six month bare fallow followed, after which maize was sown for the start of a new cycle.

The trial was set up in two phases (Table 2), so that all three crops were present across both phases in the same year from 2002 onwards. Each phase finished a cycle in two years.

| Year  | 2004 |   |  |  |   |                  | 2005 |     |   |          |  |  |  |    |                |   |     |     |                |   |   |     |
|-------|------|---|--|--|---|------------------|------|-----|---|----------|--|--|--|----|----------------|---|-----|-----|----------------|---|---|-----|
| Month |      | – |  |  | v |                  |      | . . |   | <b>.</b> |  |  |  | ۰V | $\overline{1}$ | M |     | ٠., | $\overline{1}$ |   | ◡ |     |
| Phase |      |   |  |  |   | <br>- - -<br>. . | a    |     | a |          |  |  |  |    | œ<br><b>d</b>  |   |     |     |                |   |   | . . |
| Phase |      |   |  |  |   |                  |      |     |   |          |  |  |  |    |                |   | - - |     |                | a |   |     |

Table 2 Crop rotation sequences in the two growth phases in 2004 and 2005

winter wheat  $\sum$ , soybean  $\Box$ , maize  $\Box$ , fallow  $\Box$ 

a roots sampled for the assessment of AM colonisation

There were two crop rotations started in 2001 (Phases) to allow for the three crops to be present at any one year from 2002. Phase 1: maize (April–September 2001), winter wheat (September 2001–June 2002), soybean (July–October 2002) and fallow (October 2002– April 2003). Phase 2: soybean (July–October 2001), fallow (October 2001–April 2002), maize (April–September 2002), winter wheat (September 2002–June 2003)

## Field management

Fertilisers, consisting of  $(NH4)_{2}HPO_{3}$  (diammonium phosphate, DAP, 300 kg/ha) and urea  $(CO(NH<sub>2</sub>)<sub>2</sub>$ , 150 kg/ha), were added to the winter wheat plots before sowing and at the jointing stage, respectively. The same amount of DAP was applied to maize before sowing and another 300 kg/ha of urea was applied during the growing season. Soybean received 63 kg/ha phosphorus  $(P_2O_5)$  before sowing but no extra nitrogen fertiliser was applied during the growing season. Weeds in the plots were removed by hand.

# Harvesting and sampling

Plant roots for determination of AM colonisation were collected randomly by digging four plants from each plot at an early growth stage: after winter for winter wheat; at the beginning of April, about 50 days after emergence, for maize; and 40 days after emergence for soybean. Times of sampling (see Table [2\)](#page-2-0) were chosen to obtain root material at similar developmental stages for each crop. Sampling was repeated at a late growth stage; flowering for maize and winter wheat, and pod stage for soybean. Plant roots were carefully washed free of soil, cleared in 10% KOH at 90°C in a water bath for 40–60 min according to plant species and sampling stage, dipped in 0.1 N HCl for 30 s, then stained with 0.05% trypan blue at 90°C in a water bath for 10 min (Phillips and Hayman [1970](#page-9-0)). AM colonisation was determined by the grid intersect method of Giovannetti and Mosse [\(1980\)](#page-9-0).

Four plants of winter wheat were sampled from each plot in April 2004 for isolation of root-invading fungi by the method of Nan [\(1995](#page-9-0)). Roots were washed free of soil with a jet of tap water. The washed roots were surface-sterilised in 0.3% sodium hypochlorite for 1 min, rinsed in sterile tap water, blotted dry, and then the primary tap-root was cut into 1–2 mm long segments as described by Skipp et al. ([1986\)](#page-10-0). Ten segments of each root were randomly picked and plated on Potato-Dextrose Agar (PDA) medium (Skipp et al. [1986\)](#page-10-0). In total, there were 40 segments for each plot and hence 160 segments for each treatment. Plates were incubated at 20°C in the dark until fungi could be identified or sub-cultured onto PDA for later identification. The fungi were microscopically identified based on morphological characteristics of the asexual reproductive structures and colony characters (Nan [1995](#page-9-0)). The percentage isolation for each fungus (1) and for the total rootinvading fungi (2) per root were determined as follows:

Root-invading fungus per root

 $=$  (number of segments with a particular fungus)

total number of segments)  $\times 100$ 

Total root-invading fungi per root

 $=$  (number of segments with any fungus)

total number of segments)  $\times 100$ 

Plant dry biomass was determined after crop maturity. Maize and winter wheat biomass was determined from samples from two rows (width), and 1 m (length). Soybean biomass was determined from  $0.25$  m<sup>2</sup> samples of the crop. In each case there were three replicate crop samples from each plot. Crops were chopped by hand close to the ground and seeds were later threshed by hand in a field laboratory. Grain yields of the three crops were determined after the seeds had been dried at 40°C in an oven for 48 h. The above-ground biomass of crops was dried at 85°C for 48 h, and then ground finely for analysis. P concentrations were determined colorimetrically using the phosphovanado-molybdate method (Hanson [1950](#page-9-0)) after digestion of the dried, ground plant material in a mix of concentrated perchloric and nitric acids. The data for grain yield (but not total biomass) of winter wheat and soybean in 2004 have been previously published in Chinese (Luo et al. [2005\)](#page-9-0) and are included here in order to permit analysis of the data across both years and for all crops.

# Statistical analysis

We analysed the effects of the different practices on grain yield, P uptake, above-ground biomass and AM colonisation of the three crops (in 2004 and 2005), and on the individual and total percentage isolation of root-invading fungi in winter wheat (in 2004) using analysis of variance (ANOVA). Years 2004 and 2005 overlapped with phase I and II respectively, thus we did not include Phase as source of variation. We chose <span id="page-4-0"></span>not to treat 'crop' as a factor because of the expected large differences in biomass, yield and P uptake between the different species. Similarly, we chose to analyse the data from 2004 and 2005 separately because there were large between-year differences for these parameters. We interpreted this as result of differences in complex environmental factors. Without a suitable way of discerning how all these factors and their interactions affected the results, we considered that any attempt to analyse the between-year effects and their interactions would not provide further information.

We include "residue" and "tillage" as the main factors in all the ANOVAs. "Stage" was also included in the analyses of the data from AM colonisation. Percentages were  $\sqrt{(x/100)}$  transformed before the analyses to meet the assumptions of the ANOVA (Sokal and Rohlf [1981](#page-10-0)). Means were compared using the least significant difference (LSD) method  $(P<$ 0.05). All analyses were performed with SPSS for Windows version 11.5, SPSS Inc.

# Results

Grain yield, biomass and P uptake

Grain yields from the different crops varied, with maize > winter wheat > soybean, as expected from typical yields in the region. Average yields for all crops were higher in 2005 than in 2004 (Fig. 1).

In 2004 residue application increased maize and soybean grain yield, independent of tillage treatments (CT or NT). CT slightly increased maize yield but had no effect on soybean yield. In 2005, residue application again increased soybean yield, but only under CT. A similar trend was observed in maize (Fig. 1; Table [3](#page-5-0)). There was no effect of residue application or tillage treatment on grain yield of wheat in either year (Fig. 1; Table [3](#page-5-0)).

The effects of the different agricultural practices on P uptake and above-ground biomass were similar to grain yield (see Supporting information S1 and S<sub>2</sub> and Table [3](#page-5-0)), with the exception that there was no effect of tillage on the P uptake of maize in 2004 or of soybean in 2005. Residue application increased P uptake of maize in 2004 and of soybean in 2004 and 2005. There was a trend towards increased P uptake of maize under CTR in 2005.



Fig. 1 Grain yield (t/ha) of maize, winter wheat and soybean on the Loess Plateau of China under four agricultural practices in 2004 and 2005. Bars are means of four replicates  $\pm$  standard error of the mean. CT conventional tillage (till twice, in July after harvest and September before planting), without addition of residues, NT no tillage, without addition of residues, NTR no tillage with the residue from previous crop returned to the field, CTR conventional tillage with the residue from previous crop returned to the field. Data for wheat and soybean in 2004 from Luo et al. [2005](#page-9-0), with permission. Within each crop and year, different letters show significant differences between means (maize and soybean 2005) or group of means (significant main effect of residue treatments in maize and soybean 2004); (\*) indicates significant main effect of tillage treatments in maize 2004. There were no effects of tillage or residue on winter wheat. See Table [3](#page-5-0) for details on main effects and interactions

## AM colonisation

AM colonisation of the crops was well established by the early sampling time and varied with agricultural practices and growth stage (Fig. [2](#page-6-0)). All crops had higher AM colonisation at the later sampling time than at the early stage with all agricultural practices

<span id="page-5-0"></span>

in both years of the investigation, with the exception of maize and soybean in 2004 which had the same AM colonisation at the two sampling times, and wheat in 2005 where only CTR had higher AM colonisation at later sampling time than that at the early stage. Percent root colonisation was in the range: 11 –30% (maize), 8 –31% (winter wheat) and 3 –30% (soybean).

Overall, the effect of NT on AM colonisation depended on the application of residues and the growth stage. Crops grown under NT had higher percent colonization than under CT only when no residues were added. This was consistent for all three crops at both growth stages in 2005. When residues were applied the percent colonisation at the late sampling in 2005 was higher in CTR than in NTR for all crops. This effect was apparent at the early sampling only in soybean. In 2004, the effects of tillage treatments and residue application on AM colonisation of winter wheat and soybean were also interdependent, but the pattern was not as clear as in 2005. In maize, residue application reduced the percent colonisation independent of tillage treatments. Plants grown under NT had higher percent colonisation than under CT at the early stage of growth, but the reverse was observed at the later stage.

Root-invading fungi in winter wheat

A total of 24 fungal species were isolated from roots of winter wheat and identified. The main five species of fungi isolated from all the treatments were, in order of decreasing percent isolation, Phoma medicaginis (7.1%), Ascochyta spp. (5.4%), Acremonium spp. (5.0%), Fusarium spp. (4.8%) and Rhizoctonia sp. (4.2%) (Table [4](#page-7-0)). Similar fungal species were isolated from the different treatments. No differences in total percent isolation were found among the different agricultural practices, but the trend was  $CT > NT >$ CTR > NTR (Table [4](#page-7-0)). Individual fungi showed strong responses to tillage and residue application, but there was no consistent pattern: residue application decreased the percent isolation of Ascochyta, Fusarium graminearum and Phoma medicaginis, but increased percent isolation of F. avenaceum and Cladosporium sp.. Pythium was present only in NT treatments (with or without residue application). The percent isolation of Rhizoctonia sp. was higher in NT than in CT. Values from CTR and NTR treatments were similar

Table 3 Effects of the different agricultural practices on grain yield, P uptake and biomass (ANOVA P values) Effects of the different agricultural practices on grain yield, P uptake and biomass (ANOVA P values)



<span id="page-6-0"></span>

Fig. 2 Percentage AM colonisation of maize, winter wheat and soybean under four agricultural practices at early and late stages of growth in 2004 and 2005. For treatment description see Fig. [1](#page-4-0). Bars are means of four replicates  $\pm$  standard error of the mean. Different letters indicate treatment means are significantly

and not different from NT (Table [3](#page-5-0)). No root damage or below or above ground disease symptoms were observed.

#### **Discussion**

The aim of our work was to investigate the effects of no tillage and residue application recently introduced to agriculture on the Loess Plateau on the incidence of two groups of root-invading fungi (AM fungi and pathogens) which might be expected to have positive and negative effects respectively on plant growth, and to evaluate concurrent impacts of



different within each crop and year  $(P<0.05)$ . The three-way interactions between stage, tillage and residue were significant  $(P<0.05)$  for all crops in 2004 and 2005 (data not shown) with the exception of maize 2004 where residue and tillage had independent effects (residue  $P=0.0133$ ; stage  $\times$  tillage  $P=0.026$ )

these practices on growth and yield of wheat, maize and soybean.

Grain yield, biomass and P uptake

Crop yield was generally higher in 2005 than in 2004, which can be attributed to the higher and more even distribution of rainfall in 2005 (data not shown). In 2004 rain fell for 2 weeks after the harvest of maize so that sowing of winter wheat was delayed. A similar problem occurred in 2003, also resulting in lower than average grain yields (Luo et al. [2005](#page-9-0)).

Grain yield was not significantly decreased by either NT or residue application in our study, which is

| Fungi                  | Percentage isolation | Treatment effects <sup>c</sup> |                  |                  |              |
|------------------------|----------------------|--------------------------------|------------------|------------------|--------------|
|                        | <b>CT</b>            | NT                             | <b>CTR</b>       | <b>NTR</b>       |              |
| Acremonium sp.         | $5.0a^a$             | 5.8 a                          | 8.3 a            | 0.8 <sub>b</sub> | $R \times T$ |
| Alternaria alternata   | 1.7 a                | 1.7a                           | 0.8a             | 3.3a             | none         |
| Ascochyta sp.          | 8.3 a                | 5.0a                           | 4.2 <sub>b</sub> | 4.2 <sub>b</sub> | $\mathbb{R}$ |
| Aspergillus sp.        | $N/D^b$              | 0.8a                           | 0.5a             | N/D              | $R \times T$ |
| Bipolaris sorokiniana  | N/D                  | 2.5a                           | 0.8a             | 0.8a             | none         |
| Cladosporium sp.       | N/D                  | N/D                            | 2.5a             | 3.3a             | $\mathbb{R}$ |
| Fusarium avenaceum     | N/D                  | 0.8 <sub>b</sub>               | 2.3a             | 5.0a             | $\mathbb{R}$ |
| Fusarium graminearum   | 2.5a                 | 0.8a                           | N/D              | N/D              | R            |
| <i>Fusarium</i> spp    | 5.8 a                | 5.0a                           | 5.0a             | 3.3a             | none         |
| Penicillium sp.        | N/D                  | 0.8a                           | 1.5a             | N/D              | none         |
| Phoma medicaginis      | 12.5a                | 5.8 a                          | 5.0 <sub>b</sub> | 5.0 <sub>b</sub> | $\mathbb{R}$ |
| Pythium sp.            | N/D                  | 0.8                            | N/D              | 0.8              | T            |
| Rhizoctonia sp.        | 1.7 <sub>b</sub>     | 6.7 a                          | $4.2$ ab         | $4.2$ ab         | $R \times T$ |
| Stemphylium sp.        | 2.5                  | N/D                            | N/D              | N/D              | none         |
| Thielaviopsis basicola | 1.7 <sub>b</sub>     | $4.2$ ab                       | 4.8 a            | 1.2 <sub>b</sub> | $R \times T$ |
| Ulocladium sp.         | $1.7$ ab             | 0.8 <sub>b</sub>               | 0.8 <sub>b</sub> | 3.3a             | $R \times T$ |
| total                  | 43.4a                | 41.5a                          | 40.7a            | 35.4a            | none         |

<span id="page-7-0"></span>Table 4 Mean percentage isolation of root-invading fungi isolated from winter wheat at the early growth stage under four agricultural practices.  $n=4$ 

CT conventional tillage (till twice, in July after harvest and September before planting) and no residue return

NT no tillage and no residue return

CTR conventional tillage with the residue from previous crop returned to the field

NTR no tillage with the residue from previous crop returned to the field

<sup>a</sup> Within each row, different letters indicate means are significantly different ( $P < 0.05$ )

<sup>b</sup> N/D: not detected

<sup>c</sup> Significant main effects or interactions as per ANOVA; R: residue treatments; T: tillage treatments

in agreement with previous research on this trial and strengthens the finding that modification of agricultural practices to reduce tillage and retain crop residues is unlikely to have a negative effect on crop yield in the long term (see also Luo et al. [2005](#page-9-0)). Results from another site on the Loess Plateau have also shown that although NT can result in relatively low yield of field peas, it did not decrease the average crop yield over four years in a spring wheat/field pea rotation (Huang et al. [2008](#page-9-0)). The newly introduced practices apparently carry no penalty for crop production and may even increase yields in this area, seen here in the effect of residue applications under conventional tillage (CTR). This is contrary to what has been found in some other minimum or no tillage trials, where high negative or positive effects on yield were found. For example, minimum tillage decreased crop yield by more than 40% in South Australia because of the increased severity of rhizoctonia root rot (Roget [1995\)](#page-9-0). Conversely, minimum tillage increased winter wheat yield more than 22% in Ningxia, China after ten years, the reason being that reduced tillage increased soil water storage capacity, water use efficiency and emergence rate of seedlings compared with tilled treatments (Yao et al. [2002\)](#page-10-0). The trend towards increased P uptake by maize and soybean under CTR, particularly in 2005, may reflect the influence of tillage on nutrient release from the crop residues, with consequent effects on yield.

Root-invading fungi of winter wheat

Winter wheat is the main continuous monoculture crop in the region where the trial was carried out.

Root-invading fungi can cause root rot and yield loss in some years (Nan et al., unpublished, 2003–2004). Nearly all fungal species isolated from winter wheat root are pathogenic and some of them, such as Fusarium spp. and Rhizoctonia sp., cause serious root rot and aboveground diseases (Pankhurst et al. [2002;](#page-9-0) Skoglund and Brown [1988](#page-10-0)). However, no disease symptoms were found in our study. This, and the low value of the total percentage isolation (under 50%), may be due to the low annual rainfall, low temperature in winter and/or crop rotation in the area of the trial (Bailey and Lazarovits [2003](#page-9-0)). Nevertheless, our data show that the fungi persist in all tillage and residue application treatments.

Tillage and residue application have previously been shown to suppress root rot disease caused by fungi in soil, including R. solani (Pankhurst et al. [2002\)](#page-9-0), as also shown here in the lower isolation of rhizoctonia and the absence of Pythium in CT treatments. However, total isolation of root-invading fungi did not show a significant response to tillage or residue application in our experiment, although the variation in the responses of the individual fungi may be due to differences in tolerance to tillage (Skoglund and Brown [1988\)](#page-10-0).

#### Effects on mycorrhizal colonisation

AM colonisation of all three crops was generally rather low (<30%), but differences in mycorrhizal colonisation were found among crops, tillage treatments and between years. Here, the percentage of AM colonisation was lower than in previous studies which reported values of up to 89% in wheat (Graham and Abbott [2000](#page-9-0)) and 80% in maize and soybean (McGonigle et al. [1999\)](#page-9-0). The low AM colonisation in our study may have been the result of high P fertiliser application (300 kg/ha). High P often results in low colonisation (Smith and Read [2008\)](#page-10-0) and McGonigle et al. [\(1990\)](#page-9-0) showed that when P fertiliser application was 200 kg/ha, mycorrhizal development in maize roots was significantly reduced compared with lower P applications. Colonisation of wheat roots by AM fungi has also been shown to be reduced after repeated applications of P fertilisers in soils from southern Western Australia (Graham and Abbott [2000\)](#page-9-0).

Overall no tillage (NT) alone (but not NTR) increased mycorrhizal colonisation of all the crops, which is in agreement with previous work showing that minimum or no tillage were associated with higher root colonisation in subterranean clover and maize (Jasper et al. [1989](#page-9-0); Kabir et al. [1997;](#page-9-0) McGonigle and Miller [1996](#page-9-0)), probably because tillage destroyed the hyphal network and hence reduced the infectivity of the soil.

At harvest in 2005, the influence of residue application on AM colonisation varied with tillage treatment; crops had low AM colonisation in NTR compared with both CTR and NT, which were higher for all crops (Fig. [2\)](#page-6-0). A previous study on the effect of residue application on AM colonisation has shown contradictory results (Palenzuela et al. [2002](#page-9-0)). Factors such as type and origin of the applied residue influence the activity of microorganisms in the rhizosphere, which in turn alters the AM infectivity of the soils. Furthermore, complex changes in soil chemical properties (e.g. pH, nutrient availability) are also expected (Borie et al. [2002\)](#page-9-0). The mechanisms underlying the results from the Loess Plateau require further investigation.

AM fungi may have made a contribution to P uptake by the crops, although effects were not apparent in plant P content or growth. This is expected in wheat (a non-responsive AM plant), which can show growth depression even when AM fungi contribute considerably (up to 80%) to the total plant P (Li et al. [2006](#page-9-0)). Maize and soybean have been shown to respond positively to AM colonisation when plants are grown at low levels of P (McGonigle and Miller [1996](#page-9-0); Young et al. [1986\)](#page-10-0). The lack of response of maize and soybean in our experiment might have been due to the high P fertiliser application.

It is important to note that the crop residues were not incorporated into the soil as in some other studies elsewhere. Mixing the residues with the soil can have a great impact on nutrient dynamics and hence on the effect of AM fungi on plant growth (Borie et al. [2002\)](#page-9-0).

The no tillage and residue return practices did not carry penalties in terms of reduced yield, as has been observed in similar trials elsewhere. An effect of conventional tillage in reducing AM colonisation (as shown in some previous investigations) was occasionally apparent in the presence of added residues. In general therefore, any contribution of AM symbioses to nutrient uptake and soil structural stability are likely to be maintained in the different practices. Importantly, the changed management practices did <span id="page-9-0"></span>not result in increased percent isolation of potentially pathogenic fungi, suggesting that increased disease is unlikely to be a negative outcome of these new practices in this system.

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#### References

- Bailey KL, Lazarovits G (2003) Suppressing soil-borne disease with residue management and organic amendments. Soil Tillage Res 72:169–180
- Borie F, Redel Y, Rubio R et al (2002) Interactions between crop residue application and mycorrhizal development and some soil-root interface properties and mineral acquisition by plants in an acidic soil. Biol Fertil Soils 36:151–160
- Evans DG, Miller MH (1988) Vesicular-arbuscular mycorrhizas and the soil-disturbance-induced reduction of nutrient absorption in maize. I. Causal relations. New Phytol 110:67–74
- Fairchild GL, Miller MH (1988) Vesicular-arbuscular mycorrhizas and the soil-disturbance-induced reduction of nutrient absorption in maize. II. Development of the effect. New Phytol 110:75–84
- Fu BJ (1989) Soil erosion and its control on the Loess Plateau of China. Soil Use Manage 5:76–82
- Giovannetti M, Mosse B (1980) An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots. New Phytol 84:489–500
- Graham JH, Abbott LK (2000) Wheat responses to aggressive and non-aggressive arbuscular mycorrhizal fungi. Plant Soil 220:207–218
- Hanson WC (1950) The photometric determination of phosphorus in fertilizers using the phosphovanado-molybdate complex. J Sci Food Agric 1:172–173
- Hardiman RT, Lacey R, Yang MY (1990) Use of cluster analysis for identification and classification of farming systems in Qingyang County, central North China. Agric Syst 33:115–125
- Huang GB, Zhang RZ, Li GD et al (2008) Productivity and sustainability of a spring wheat-field pea rotation in a

semi-arid environment under conventional and conservation tillage systems. Field Crop Res 107:43–55

- Jasper DA, Abbott LK, Robson AD (1989) Soil disturbance reduces the infectivity of external hyphae of vesiculararbuscular mycorrhizal fungi. New Phytol 112:93–99
- Kabir Z, O'Halloran IP, Fyles JW et al (1997) Seasonal changes of arbuscular mycorrhizal fungi as affected by tillage practices and fertilization: hyphal density and mycorrhizal root colonisation. Plant Soil 192:285–293
- Li HY, Smith SE, Holloway RE et al (2006) Arbuscular mycorrhizal fungi contribute to phosphorus uptake by wheat grown in a phosphorus-fixing soil even in the absence of positive growth responses. New Phytol 172:536–543
- Liu GB (1999) Soil conservation and sustainable agriculture on the Loess Plateau: challenges and prospects. Ambio 28:663–668
- Luo CY, Shen YY, Nan ZB et al (2005) Dynamics of crop yield and soil oxidizible organic carbon fraction within a maizewinter wheat-soybean rotation under different tillage treatment in Longdong Loess Plateau. J Soil Water Conserv 19:84–88 (In Chinese)
- McGonigle TP, Miller MH (1996) Mycorrhizae, phosphorus absorption, and yield of maize in response to tillage. Soil Sci Soc Am J 60:1856–1861
- McGonigle TP, Evans DG, Miller MH (1990) Effect of degree of soil disturbance on mycorrhizal colonisation and phosphorus absorption by maize in growth chamber and field experiments. New Phytol 116:629–636
- McGonigle TP, Miller MH, Young D (1999) Mycorrhizae, crop growth, and crop phosphorus nutrition in maize-soybean rotation given various tillage treatments. Plant Soil 210:33– 42
- Nan ZB (1995) Fungicide seed treatments of sainfoil control seed-borne and root-invading fungi. New Zeal J Agric Res 38:413–420
- Olsen SR, Cole CV, Watanabe FS, Dean LA (1954) Estimation of available phosphorus in soils by extraction with sodium bicarbonate. Circular 939. United States Department of Agriculture, Washington, D.C.
- Palenzuela J, Azcon-Aguilar C, Figueroa D et al (2002) Effects of mycorrhizal inoculation of shrubs from Mediterranean ecosystems and composted residue application on transplant performance and mycorrhizal development in desertified soil. Biol Fertil Soils 36:170–175
- Pankhurst CE, McDonald HJ, Hawke BG et al (2002) Effect of tillage and stubble management on chemical and microbiological properties and the development of suppression towards cereal root disease in soils from two sites in NSW, Australia. Soil Biol Biochem 34:833–840
- Phillips JM, Hayman DS (1970) Improved procedures for clearing roots and staining parasitic and vesiculararbuscular mycorrhizal fungi for rapid assessment of infection. Trans Br Mycol Soc 55:158–161
- Roget DK (1995) Decline in root rot (Rhizoctonia solani AG-8) in wheat in a tillage and rotation experiment at Avon, South Australia. Aust J Exp Agric 35:1009–1013
- Rovira AD (1986) Influence of crop rotation and tillage on Rhizoctonia Bare Patch of Wheat. Phytopathology 76:669–673
- <span id="page-10-0"></span>Shi H, Shao MG (2000) Soil and water loss from the Loess Plateau in China. J Arid Environ 45:9–20
- Skipp RA, Christensen MJ, Nan ZB (1986) Invasion of red clover (Trifolium pratense) roots by soil-borne fungi. New Zeal J Agric Res 29:305–313
- Skoglund LG, Brown WM (1988) Effects of tillage regimes and herbicides on Fusarium species associated with corn stalk rot. Can J Plant Pathol 10:332–338
- Smith SE, Read DJ (2008) Mycorrhizal symbiosis, 3rd edn. Academic, London
- Sokal RR, Rohlf FJ (1981) Biometry, 2nd edn. WH Freeman and Company, New York
- Tisdall JM, Oades JM (1979) Stabilization of soil aggregates by the root systems of ryegrass. Aust J Soil Res 17:429–441
- Triplett Jr GB, Dick WA (2008) No-tillage crop production: a revolution in agriculture! In: Celebrate the Centennial, A supplement to Agron J 100:S153-S165
- Yao YQ, Wang YH, Lv JJ (2002) Water dynamics and soil erosion under conservation wheat field. Soil Fertil 5:8–10 (In Chinese)
- Young CC, Juang TC, Guo HY (1986) The effect of inoculation with vesicular-arbuscular mycorrhizal fungi on soybean yield and mineral phosphorus utilization in subtropicaltropical soils. Plant Soil 95:245–253